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Veterinary Medicines Division

Committee for Medicinal Products for Veterinary Use

CVMP assessment report for CircoMax Myco (EMEA/V/C/005184/0000)

Vaccine common name: Porcine circovirus vaccine (inactivated, recombinant) and *Mycoplasma hyopneumoniae* vaccine (inactivated)

Assessment report as adopted by the CVMP with all information of a commercially confidential nature deleted.

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Introduction

The applicant Zoetis Belgium SA submitted on 6 September 2019 an application for a marketing authorisation to the European Medicines Agency (the Agency) for CircoMax Myco, through the centralised procedure under Article 3(1) of Regulation (EC) No 726/2004 (mandatory scope).

The eligibility to the centralised procedure was agreed upon by the CVMP on 11 October 2018 as the PCV antigens in CircoMax Myco have been developed using recombinant DNA technology.

The applicant applied for the following indication:

- Active immunisation of pigs over the age of 3 days against porcine circovirus type 2 to reduce viral load in blood and lymphoid tissues, virus fecal shedding and the lesions in lymphoid tissues associated with PCV2 infection.
- Active immunisation of pigs over the age of 3 days against *Mycoplasma hyopneumoniae* to reduce the lung lesions associated with *Mycoplasma hyopneumoniae* infection.
- To reduce the loss of weight gain during the finishing period in the face of infection with PCV2 (as observed in field studies).

  Onset of immunity (both vaccination schedules): from 3 weeks after (the last) vaccination.

  Duration of immunity (both vaccination schedules): 23 weeks after (the last) vaccination.

The final proposal for the indications is:

Active immunisation of pigs against porcine circovirus type 2 to reduce viral load in blood and lymphoid tissues, fecal shedding and the lesions in lymphoid tissues associated with PCV2 infection. Protection was demonstrated against porcine circovirus genotypes 2a, 2b and 2d.

Active immunisation of pigs against *Mycoplasma hyopneumoniae* to reduce the lung lesions associated with *Mycoplasma hyopneumoniae* infection.

Onset of immunity (both vaccination schedules): 3 weeks after (the last) vaccination.

Duration of immunity (both vaccination schedules): 23 weeks after (the last) vaccination.

In addition, vaccination has been shown to reduce body weight gain losses under field conditions

The active substances of CircoMax Myco are inactivated recombinant chimeric porcine circovirus 1 containing the porcine circovirus 2a open reading frame 2 (ORF2) protein, inactivated recombinant chimeric porcine circovirus 1 containing the porcine circovirus 2b ORF2 protein and inactivated *Mycoplasma hyopneumoniae* (MH). CircoMax Myco is a trivalent, inactivated, emulsified vaccine to induce immunity against PCV2 and *M. hyopneumoniae*. The target species are pigs for fattening. The product is intended for administration by intramuscular use.

CircoMax Myco emulsion for injection contains 1.5 – 4.9 relative potency units (RP) of inactivated recombinant chimeric porcine circovirus type 1 containing the porcine circovirus type 2a ORF2 protein, 1.5 – 5.9 RP of inactivated recombinant chimeric porcine circovirus type 1 containing the porcine circovirus type 2b ORF2 protein and 1.5 – 4.7 RP of inactivated *Mycoplasma hyopneumoniae*, strain P-5722-3 and is presented in the following packs:

- Cardboard box of 1 vial of 50 ml (25 doses), 100 ml (50 doses) or 250 ml (125 doses).
- Cardboard box of 10 vials of 50 ml (25 doses) or 100 ml (50 doses).
- Cardboard box of 4 vials of 250 ml (125 doses).

The rapporteur appointed is Niels Christian Kyvsgaard and the co-rapporteur is Paolo Pasquali.

The dossier has been submitted in line with the requirements for submissions under Article 12(3) of Directive 2001/82/EC – full application.

Marketing authorisation under exceptional circumstances

Not applicable.

Scientific advice

Not applicable.

MUMS/limited market status

Not applicable.

Multi-strain dossier

Not applicable.

Part 1 - Administrative particulars

Detailed description of the pharmacovigilance system

A detailed description of the pharmacovigilance system (dated 28 May 2018) which fulfils the requirements of Directive 2001/82/EC was provided. Based on the information provided the applicant has the services of a qualified person responsible for pharmacovigilance and the necessary means for the notification of any adverse reaction occurring either in the Community or in a third country.

Manufacturing authorisations and inspection status

Manufacture of the active substances and final product takes place outside the EEA at Zoetis Inc Iowa, United States, and Zoetis Inc Nebraska, United States. The sites have a manufacturing authorisation issued on 03 October 2018 by Unites States Department of Agriculture. GMP certifications, which confirms the dates of the last inspections and show that the sites are authorised for the manufacture and batch release of such veterinary dosage forms, were issued by the Veterinary Medicines Directorate, United Kingdom.

Secondary packaging and batch release within the EU take place at Zoetis Belgium SA, Louvain-la-Neuve, Belgium which holds a manufacturing authorisation issued on 03 March 2018 by the Federal Agency for Medicines and Health Products, BE. GMP compliance was confirmed by the competent national authority Federal Agency for Medicines and Health Products, BE.

Overall conclusions on administrative particulars

The detailed description of the pharmacovigilance system was considered in line with legal requirements.

The GMP status of the active substance(s) and of the finished product manufacturing sites has been satisfactorily established and are in line with legal requirements.

Part 2 – Quality

Chemical, pharmaceutical and biological/microbiological information (quality)

Qualitative and quantitative particulars of the constituents

Qualitative and quantitative particulars

CircoMax Myco is a trivalent inactivated vaccine intended for active immunisation of pigs over the age of three days to reduce virus load, virus shedding, and lesions associated with Porcine circovirus type 2 infection and lung lesions of *Mycoplasma hyopneumoniae* infection.

The finished product is an emulsion for injection. The active substances (antigens) are inactivated *Mycoplasma hyopneumoniae* strain P-5722-3 (MH), inactivated recombinant chimeric porcine circovirus (PCV) type 1 containing the PCV type 2a ORF2 protein (PCV2a), and inactivated recombinant chimeric PCV type 1 containing the PCV type 2b ORF2 protein (PCV2b). The product contains (in relative potency (RP) units): MH, 1.5-4.7 RP; PCV2a, 1.5-4.9 RP; and PCV2b, 1.5-5.9 RP per dose of 2 ml. The product contains squalane (as SP oil solution, also known as Metastim®, which in addition to squalane contains poloxamer 401 and polysorbate 80) as adjuvant. Other ingredients are EDTA tetrasodium, disodium tetraborate decahydrate, buffer salts (monobasic potassium phosphate, disodium phosphate, sodium phosphate dibasic), sodium chloride, and potassium chloride as described in section 6.1 of the SPC. Thiomersal is added as preservative.

CircoMax Myco is available in multidose high density polyethylene (HDPE) plastic vials containing 25, 50, or 125 doses of 2 ml, closed with pharmaceutical-grade chlorobutyl rubber stoppers and sealed with aluminium caps, as described in section 6.5 of the SPC.

Overall, the composition of the vaccine is adequately described.

Container and closure

CircoMax Myco is filled into multidose HDPE plastic vials (50, 100, or 250 ml), containing 25, 50, or 125 doses of 2 ml, respectively. The vials are closed with chlorobutyl rubber stoppers (Ph. Eur. 3.2.9) and sealed with aluminium caps. HDPE vials are sterilised by gamma irradiation at ≥ 25 kGy, according to Ph. Eur. 5.1.1. Rubber stoppers are steam sterilised (Ph. Eur. 5.1.1). Certificates of analysis have been supplied for containers and closure demonstrating compliance with the proposed specifications.

Product development

CircoMax Myco is based on the predecessor product Suvaxyn Circo + MH RTU approved in EU; CircoMax Myco is a new vaccine containing the additional antigen PCV2b, the manufacture of which is based on the same technology as for the PCV antigen in the parent product.

The new additional component PCV2b antigen was obtained by recombinant technology using the same porcine circovirus type 1 vector strain used as backbone for the PCV2a antigen, producing a chimera expressing porcine circovirus type 2b capsid protein. As for PCV2a antigen, in this construct the PCV2b capsid gene (ORF2) itself is avirulent and non-infectious, and the PCV1 genome backbone is non-pathogenic. By including the PCV2b antigen a broader immune coverage is provided against current and emerging strains of PCV2 compared to vaccines based on a single PCV2 type only. The PCV2a antigen and the *M. hyopneumoniae* antigen included in CircoMax Myco are identical to the antigens used in Suvaxyn Circo + MH RTU.

The PCV2a antigen production process is identical to the one used for the licenced Suvaxyn Circo + MH RTU and the master seed virus is identical. For PCV2b antigen, a new master seed virus has been qualified, but otherwise the production process of PCV2b antigen is identical to that of PCV2a. The *M. hyopneumoniae* antigen production process is the same as for the Suvaxyn Circo + MH RTU and the master seed bacteria are the same. The inactivation procedures are the same as those used for the latter licensed vaccine.

Inactivation of *M. hyopneumoniae* is performed using Binary ethylenimine (BEI), which has been demonstrated to be capable of inactivating the proposed maximum pre-inactivation titre for *M. hyopneumoniae* (3.6×10^{10} 50% color changing units (CCU_{50})/ml), within less than 67 percent of the proposed duration of the inactivation process. This is in accordance with the requirements of Ph. Eur. 0062 *Vaccines for veterinary use*. BEI is neutralised with sodium thiosulfate at the end of the inactivation period. Control for inactivation (residual live mycoplasma) is carried out on each batch of antigen directly after the inactivation step, using a validated method (incubation in growth medium).

Inactivation of the PCV2a and PCV2b is performed by addition of Beta Propiolactone (BPL). Kinetics of inactivation studies demonstrated that the proposed inactivation procedures are capable of inactivating the proposed maximum pre-inactivation titre for PCV2a antigen (9.3 $\log_{10}$ fluorescent assay infectious dose 50% ($FAID_{50}$)/ml) and for PCV2b antigen (8.6 $\log_{10}$ $FAID_{50}$/ml) within less than 67 percent of the proposed duration of the inactivation process, in accordance with the requirements of Ph. Eur. 0062. Inactivation is terminated by addition of sodium thiosulfate after which testing for complete inactivation of virus is performed. Completeness of virus inactivation is carried out on each batch directly after inactivation by staining virus-inoculated cells with a monoclonal antibody specific for PCV2 using a validated method.

The upper and lower specifications for relative potency for each antigen are based on safety and efficacy studies. Batches used in clinical trials are listed and batch records are provided for most of the batches. The formulation of batches used during clinical studies is the same as that intended for marketing. The minimum potency release specifications were based on analyses of decrease of potency in the stability batches. Filling volume overage has been stated and justified.

All excipients are well known pharmaceutical ingredients and their quality is compliant with Ph. Eur. or internal standards. There are no novel excipients used in the finished product formulation. The list of excipients is included in section 6.1 of the SPC. Thiomersal is added as preservative. A recommendation is put forward on removal of thiomersal from the presentation.

Description of the manufacturing method

***M. hyopneumoniae* antigen**

The *M. hyopneumoniae* antigen production process is the same as for *M. hyopneumoniae* used in the approved vaccine Suvaxyn Circo + MH RTU. The manufacture of *M. hyopneumoniae* antigen is a standard

aerobic fermentation in bioreactors, followed by inactivation, purification, and sterilisation by filtration. The manufacturing is based on a seed lot system.

M. hyopneumoniae antigen is produced in fermentors with a working volume of approx. 2400 L. The scale up/fermentation process consists of four steps: inoculation and expansion of master or working seed, serial scale-up in culture vessels, serial scale-up in seed fermentors, and production in production fermentor. Inoculation volumes, cultivation times, and target temperatures are defined for each step, and approximate volumes are stated for the individual steps. pH and aeration are controlled in the production fermentor. Antifoam may be added to the production fermentor. Maximum *M. hyopneumoniae* passage level for antigen production is defined. After fermentation, the *M. hyopneumoniae* cells are inactivated by treatment with Binary Ethylenimine (BEI), completeness of inactivation is tested, and residual BEI is neutralised with sodium thiosulfate. The neutralised antigen fluid may be stored at +2 to +8 °C for 312 hours. If fluids are held for more than 24 hours, a single 0.45 µm filtration may be performed in case a precipitate appears during storage. Neutralised antigen fluid is clarified by filtration; the clarified antigen fluid may be stored at +2 to +8 °C for 336 hours. The antigen fluid is further purified by protein A chromatography. Finally, the antigen is sterilised by filtration. The final antigen fluid is stored in polyethylene bags (or equivalent) at +2 to +8 °C; the shelf life is 2 years. The typical batch size of the final *M. hyopneumoniae* antigen fluids is 400-900 L.

PCV2a and PCV2b antigens

The PCV2a antigen production process is identical to the cPCV1-2 antigen used in the approved vaccine Suvaxyn Circo + MH RTU, and the manufacturing process of the new PCV2b antigen is identical to that of PCV2a. The manufacturing process is a standard virus production in pig kidney PK-15 cells followed by separation of micro carriers and cell material, and concentration, purification, and inactivation of virus. The manufacturing process is based on a seed lot system.

The production process consists of the following steps: scale up of WCS of PK-15 cells, inoculation of cells with PCV seed, and production cultivation on microcarriers in bioreactors. Multiplicity of infection is defined, and cultivation times and target temperatures are set for the cultivation steps. Gentamycin is used during the cultivation steps. Culture fluid containing both cell-free virions and virus associated to infected cells that have detached from the microcarriers as a result of virus infection, is harvested. Harvest of viral fluids and subsequent replacement with fresh growth media may be accomplished repeatedly and up to four harvests may be collected. Physical agitation may be used to assist in removal of cells from the microcarriers. Initial harvest is based on visual observation of cultures and/or evaluation of bioreactor control parameters *e.g.* dissolved oxygen consumption. The maximum cumulative incubation time from inoculation with frozen seed to final harvest is defined. The maximum passage levels are defined for cells and virus and verified by genetic stability studies. Harvest is followed by removal of microcarriers by screen filtration, concentration by filtration (the harvest stocks may be combined before concentration), and purification by diafiltration. The concentrated antigen stocks can be stored up to 30 days at +2 to +8 °C prior to inactivation. Inactivation of viral fluid is performed by addition of Beta Propiolactone and terminated by addition of sodium thiosulfate after which testing for complete inactivation of virus is performed. The final antigen fluids are stored in polypropylene bags or stainless-steel vessels at +2 to +8 °C; the shelf life is 24 months. The final antigen batch sizes range between 50 and 270 L.

Vaccine Production

Bulk vaccine formulation is done by mixing the appropriate amounts of *M. hyopneumoniae*, PCV2a, and PCV2b antigens, SP-oil adjuvant, buffer salts, and thiomersal solution. The amounts of antigens added are targeted to meet the release specifications. Details of blending are provided. pH of the final bulk is controlled and adjusted if necessary. The formulated blend may be stored at +2 to +8 °C for up to 23 days. The final bulk is filtered through a coarse screen and filled into sterile multidose HDPE vials (25, 50,

or 125 doses). The filled vials are closed with the stopper and crimped. The finished product is stored at +2 to +8 °C.

Validation of the manufacturing process is demonstrated by provision of results from six batches of *M. hyopneumoniae* antigens, three batches of each of the PCV antigens (2a and 2b), and six consecutive finished product batches. The finished product batches were based on three final bulks each manufactured with different antigen batches; they were filled in 25-doses and 125-doses vials to support also the 50-doses vial presentation. It has been demonstrated that the manufacturing process is capable of producing the finished product of intended quality in a reproducible and consistent manner. The in-process controls are considered adequate for this type of manufacturing process.

Production and control of starting materials

Starting materials listed in pharmacopoeias

Starting materials listed in a pharmacopoeia are presented. The starting materials include cysteine hydrochloride monohydrate, glucose (anhydrous), glycerol, hydrochloric acid (concentrated), purified water, sodium chloride, sodium hydroxide, sodium thiosulfate, gentamicin sulphate, phenol red sodium salt, potassium chloride, potassium phosphate monobasic anhydrous, sodium hydrogen carbonate, sodium phosphate dibasic anhydrous, sodium phosphate dibasic heptahydrate, sodium dihydrogen phosphate monohydrate, disodium tetraborate decahydrate, polysorbate 80, thiomersal, water for injections, foetal bovine serum and squalane. The quality of the materials must comply with Ph. Eur with the exception of sodium phosphate dibasic anhydrous and sodium phosphate dibasic heptahydrate, which comply with USP, and phenol red, which complies with American Chemical Society (ACS). This is acceptable, as no Ph. Eur monographs exist. Examples of certificates of analyses are provided for all the listed starting materials.

Starting materials of biological (animal) origin includes fetal bovine serum and squalane (shark origin). For bovine serum, valid European Directorate for the Quality of Medicines (EDQM) certificates of suitability are provided for each of the suppliers.

Specific materials not listed in a pharmacopoeia

Starting materials of biological origin

Master and working seeds

M. hyopneumoniae strain P-5722-3:

M. hyopneumoniae strain P-5722-3 is already the basis of the authorised vaccine Suvaxyn Circo + MH RTU. The source and history of the strain is described in sufficient detail. The manufacture, testing, and storage of the master seed (MS) is adequately described and documented. The MS was tested for: Purity (sterility, as per Ph. Eur 2.6.1); Identity by i) purity (growth on agar), ii) biochemical characterisation, iii) Fluorescent antibody identity, iv) typing via 16S rDNA sequence analysis, v) qPCR amplification; Potency (color-changing unit titration); Extraneous virus (testing on NL-ST-1 porcine cells). The manufacture, testing, and storage of new working seeds (WS) is described. New working seed lots are tested for identity (Fluorescent antibody identity), purity (phase contrast microscopy), sterility (Ph. Eur 2.6.1), and titer (color-changing unit titration).

Overall, the tests performed on the master seeds and working seeds are appropriate and in accordance with Ph. Eur. 0062*, Vaccines for veterinary use*.

PCV2a:

The PCV2a chimera is already used as one of the components in the authorised vaccine Suvaxyn Circo + MH RTU. As such, the master seed virus Lot # 2117-63-052004 has already been found suitable and approved for use in the manufacture of veterinary viral vaccines. The strategy for generation and testing of future working virus seed (WSV) is included and is considered acceptable.

PCV2b:

The PCV2b chimeric virus was constructed by replacing the ORF2 of the non-pathogenic PCV1 virus backbone with the ORF2 of the pathogenic PCV type 2b by the use of fusion PCR methodology. The phenotype of the chimeric virus was verified by immunofluorescence staining of inoculated PK-15 cells and the genotype was verified by full-length sequencing. Furthermore, the genetic sequence was verified by restriction enzyme analysis.

Prior to generation of the master virus seed (MSV), the chimeric virus was subjected to three rounds of cloning via limiting dilution in PK-15 cells and several passages on cells for expansion.

The procedure for generating the pre-MSV, the MSV and the generation of future WSVs has been adequately described. The MSV has been subjected to comprehensive testing according to Ph. Eur., USP, and Annex 2 of EMA/CVMP/IWP/206555/2010-Rev.1, including testing for identity, genetic stability, and absence of bacteria, fungi, mycoplasma and a large panel of relevant viruses.

Strategy for testing of future WSVs is included and is considered acceptable.

PK-15 cell line:

The PK-15 cell line is already used for the manufacture of the PCV2a antigen, which is one of the components in the authorised vaccine Suvaxyn Circo + MH RTU. Overall testing of the MCS is in accordance with the current version of Ph. Eur. 5.2.4 *Cell cultures for the production of veterinary vaccines* as well as EMA/CVMP/IWP/206555/2010/Rev.1. For the PK-15 working cell seed (WCS), the testing is in accordance with current guidelines.

Other starting materials of biological origin

Components of porcine, bovine, and *E. coli* origin, and yeast extract (*S. cerevisiae*) are used for production of seed material and/or during fermentation of pre-cultures and main culture. The starting materials include: Porcine origin: microcarriers (contains gelatin), PPLO broth, porcine serum (heat treated, irradiated), porcine trypsin powder; Bovine origin: Lactalbumin Hydrolysate (LAH) and OptiMEM 50x Salts II Solution (contains transferrin); and *E. coli* origin: MabSelect Protein A Resin. Raw material specifications, including source and accepted countries of origin for materials of biological origin, and examples of certificates of analyses are provided for most of the listed starting materials.

The animal-based materials are considered in compliance with the requirements of Ph. Eur. 5.2.5 *'Substances of animal origin for the production of immunological veterinary medicinal products (IVMP)'.* Starting materials from transmissible spongiform encephalopathies (TSE)-relevant species include fetal bovine serum (FBS), transferrin (derived from bovine blood), lactalbumin hydrolysate (LAH, derived from bovine milk), and porcine trypsin powder (containing lactose derived from bovine milk). The bovine milk used for manufacture of LAH and trypsin is considered fit for human consumption. Valid EDQM certificates are provided for bovine serum and transferrin. The risk of transmitting TSE is considered negligible.

Starting materials of non-biological origin

SAG 471, Betapropiolactone (BPL), EMEM with phenol red, OptiMEM 50x Acid Soluble Solution, OptiMEM 50x Salts, Poloxamer 401 (adjuvant component, surfactant), sodium phosphate heptahydrate, sodium phosphate monobasic monohydrate, EDTA tetrasodium salt dehydrate, and microcarriers. Examples of certificates of analyses are provided.

In-house preparation of media and solutions consisting of several components

Media and solutions include: antifoam solution, cysteine chloride solution, anhydrous dextrose solution, PPLO broth porcine, yeast extract solution, sodium hydroxide solution, hydrochloric acid solution, equilibration buffer, sodium thiosulfate 25% solution, binary ethylene amine (BEA) 1M solution, PPLO complete medium, OptiMEM with 0.25% LAH, OptiMEM with 0.25% LAH/FBS/gentamicin, Eagle's medium with 0.05% LAH, 0.01M, Phosphate Buffer Saline (PBS), gentamicin sulphate solution, 1M sodium thiosulfate solution, 10X trypsin solution with EDTA, thiomersal 5% with EDTA and Borate, SP oil solution, homogenized SP oil solution and thiomersal 10% solution. Information on the qualitative and quantitative composition, methods of preparation, sterilisation, and storage conditions of the media and solutions is included in the dossier.

Control tests during the manufacturing process

M. hyopneumoniae antigen

The in-process control tests performed during the production of *M. hyopneumoniae* antigen are the same as those already approved for the vaccine Suvaxyn Circo + MH RTU. Descriptions of test method, and their acceptance criteria, are provided for all methods. In-process controls include analyses of purity, antigen titre prior to inactivation, completeness of inactivation, content of sodium thiosulfate, and bioburden level prior to bioburden reduction. The final antigen fluid is tested for sterility, antigen content, and content of swine IgG and protein A. The applicant presented in-process data for the manufacture of six batches of *M. hyopneumoniae* antigens. The analytical data and results are provided and are all within specifications.

Enumeration of *M. hyopneumoniae* prior to inactivation is performed by colour changing units (CCU) titration assay; an upper limit of antigen titre prior to inactivation is defined. For control of inactivation (residual live mycoplasma), samples of the inactivated culture are incubated in growth medium; control for inactivation is carried out on each batch of antigen directly after the inactivation step. The test methods are adequately validated.

Antigen content in final antigen fluid is determined by ELISA using a monoclonal antibody specific for *M. hyopneumoniae* P46 antigen as capture antibody and a peroxidase conjugate of a second monoclonal antibody as detector antibody. The antigenic content is expressed as relative units per ml compared to a reference antigen. An in-house standard is used as positive control. The ELISA method is the same as the method validated for the approved vaccine Suvaxyn Circo + MH RTU.

Swine IgG and protein A are measured by commercially available ELISA kits. Both test methods are validated as appropriate. Sterility testing is performed by membrane filtration as per Ph. Eur 2.6.1, and test of method suitability was performed.

PCV2 antigens

The analytical methods used to test the PCV2a antigen are identical to those used in the already approved vaccine Suvaxyn Circo + MH RTU. In-process controls include analyses of virus titre and identity, sterility, complete inactivation, potency, residual sodium thiosulfate, and relative purity (calculated). Descriptions of test method are provided for all methods.

Virus titre and identity before inactivation is determined by staining virus-inoculated NL-ST cells with a monoclonal antibody specific for PCV2. Upper limits of PCV2a and PCV2b titres prior to inactivation are defined. The method has been validated for both PCV2a and PCV2b. Completeness of virus inactivation is tested by staining virus-inoculated NL-ST cells. The method has been validated for PCV2a antigen. Specific validation for PCV2b antigen is not considered necessary.

The virus content in the inactivated final antigen fluid is determined by ELISA using rabbit anti PCV2 serum as capture antibody, monoclonal PCV2 antibody (5D5-5H4) for detection together with a peroxidase-conjugated secondary goat-anti-mouse antibody. In-house reference vaccine and placebo vaccine are used as controls. Validation of the ELISA procedure has been conducted for the PCV2a antigen. Internal verifications have been conducted and confirmed that the same test (and specifications) is applicable to the PCV2b antigen.

Sterility testing is performed as per Ph. Eur 2.6.1 and has been validated for method suitability.

Control tests on the finished product

Control of finished product includes analyses of general characteristics (description, pH and viscosity), potency/identification of the individual antigens, thiomersal content, quantification of adjuvant (squalane) and sterility. In addition, protein A content is calculated. Test for inactivation is not performed for batch release; this is acceptable since it is performed for the individual antigens after inactivation. Overall, the selection of parameters in the finished product specification covers the aspects that would be expected. Descriptions of test method are provided for all methods.

Potency/identification, M. hyopneumoniae:

The *M. hyopneumoniae* content of the finished product is determined by ELISA using a monoclonal antibody specific for the p46 protein content of *M. hyopneumoniae* and a monoclonal antibody labelled with peroxidase for detection. A placebo vaccine without *M. hyopneumoniae* antigen and a positive control vaccine batch are used as controls. The ELISA method is the same method as that already registered for the vaccine Suvaxyn Circo + MH RTU, the only difference being the batch of reference vaccine used and the samples to be tested (trivalent vaccine). The use of different reference vaccines in the *M. hyopneumoniae* potency test for the two products (CircoMax Myco and Suvaxyn Circo+MH RTU) makes irrelevant any direct comparison between the RPs of the two products. No formal re-validation was performed for the present trivalent vaccine, but additional experiments were conducted to confirm suitability of the assay for testing the *M. hyopneumoniae* component in the trivalent vaccine. Although the validation is minimal with respect to both samples and parameters evaluated, the validity of the ELISA assay for testing the *M. hyopneumoniae* component of the vaccine is overall considered demonstrated.

Potency/identification, PCV2a:

The PCV2a content of the finished product is determined by ELISA using a monoclonal antibody specific for PCV2a as capture antibody and a biotinylated monoclonal PCV2 antibody and commercial streptavidin peroxidase for detection. The reference vaccine is containing only *M. hyopneumoniae* and PCV2a antigens. A placebo vaccine without PCV2a antigen and a positive control vaccine batch are used as controls. The ELISA method is different compared to the ELISA method registered for Suvaxyn Circo + MH RTU. This makes irrelevant any direct comparison between the RPs of the two products. The assay is

considered validated and in line with the expectations of VICH GL1 and GL2. Overall, it is considered demonstrated that the PCV2a ELISA for finished product is able to reflect the amount of the antigen in the final product with adequate accuracy.

Potency/identification, PCV2b:

The PCV2b content of the finished product is determined by ELISA using a monoclonal antibody specific for PCV2b as capture antibody and a biotinylated monoclonal PCV2 antibody and streptavidin peroxidase for detection. The reference vaccine is containing all three antigens. A placebo vaccine without PCV2b antigen and a positive control vaccine batch are used as controls. The assay is considered validated and in line with the expectations of VICH GL1 and GL2. Overall, it is considered demonstrated that the PCV2b ELISA for finished product is able to reflect the amount of the antigen in the final product with adequate accuracy.

Sterility testing is performed as per Ph. Eur 2.6.1 and has been validated for method suitability. Determination of thiomersal is performed in accordance with the direct calibration method described in Ph. Eur. 2.2.23; the method is considered adequately validated. Determination of the squalane is based on HPLC; the method is considered adequately validated. Protein A content is calculated.

Batch-to-batch consistency

The applicant presented batch data for six batches of *M. hyopneumoniae* antigens, three batches of each of the PCV antigens (2a and 2b), and six consecutive finished product batches. The finished product batches were based on three final bulks each manufactured with different antigen batches and were filled in 25- doses and 125-doses vials to support also the 50-doses vial presentation. In general, the batch results submitted fulfil the proposed in-process control specifications and specifications for finished product and demonstrate acceptable consistency of manufacturing process and final product.

Stability

Antigens

M. hyopneumoniae antigen

The stability testing of the *M. hyopneumoniae* antigen include testing of antigen content only; stability of this parameter is considered demonstrated for the proposed shelf life of 24 months at +2 to +8 °C. Sterility of the antigen after long-term storage has been adequately demonstrated and the absence of routine sterility testing of the antigen is considered justified.

PCV2a antigen

The stability testing of the PCV2a antigen include testing of potency only; stability of this parameter is considered demonstrated for the proposed shelf life of 24 months at +2 to +8 °C. Sterility of the antigen after long-term storage has been adequately demonstrated and the absence of routine sterility testing of the antigen is considered justified.

PCV2b antigen

The stability testing of the PCV2b antigen include testing of potency and sterility. Sterility testing of the PCV2b antigen is not due to concerns regarding antigen sterility, but the PCV2b antigen stability protocol included sterility testing to support other international marketing authorizations. The results provided support the proposed shelf life of 24 months at +2 to +8 °C.

Finished product

The proposed shelf life of finished product is 18 months at +2 to +8 °C. Twenty-nine months stability data are provided for six finished product batches, three batches of 25-doses and three batches of 125 dose-vials, based on three different final bulks. The 50-dose presentation is considered supported by the bracketing approach.

The general expectations for stability testing frequency are not met, as the testing was put on hold between the 6 to 15-month time points. However, the delay in testing is overall considered acceptable and of no significant impact on the assessment of stability.

The proposed shelf life is considered supported by the stability data presented.

In-use shelf life

No in-use shelf life is claimed as the product is to be used immediately after first broaching.

Efficacy of preservative

The thiomersal content specifications are the same as those found for the licensed vaccine Suvaxyn Circo + MH RTU. The effectiveness of thiomersal was demonstrated for the Suvaxyn Circo + MH RTU vaccine in compliance with Ph. Eur. 5.1.3 and 0062. This is considered adequate and in line with *Notice to Applicants, EudraLex Volume 6B*.

Overall conclusions on quality

CircoMax Myco is a trivalent inactivated vaccine intended for active immunisation of pigs over the age of three days. The finished product is an emulsion for injection presented in HDPE vials containing 25, 50 or 125 doses. The active substances (antigens) are inactivated *Mycoplasma hyopneumoniae*, inactivated recombinant chimeric porcine circovirus type PCV2a and inactivated recombinant chimeric porcine circovirus type PCV2b.

Overall, information on the development, manufacture and control of the active substances and the finished product has been presented in a satisfactory manner.

The manufacturing of *M. hyopneumoniae* antigen is based on a seed lot system, a standard aerobic fermentation in bioreactors, followed by inactivation, purification, and sterilisation by filtration. The manufacturing processes of the two PCV antigens are identical and comprise standard virus production in pig kidney PK-15 cells followed by separation of microcarriers and cell material, and concentration, purification, and inactivation of virus. The manufacturing process is based on a seed lot system. After the applicant's acceptance of the CVMP recommendations to reduce the level of the microbial load during production of the *M. hyopneumoniae* antigen, the manufacturing processes are generally considered adequately controlled. Based on the data from six consecutive finished product batches, acceptable batch-to-batch consistency is considered demonstrated. Compliance with Ph. Eur. monographs 0062 *Vaccines for veterinary use* is generally considered demonstrated.

Data from stability studies for six batches of the finished product indicate that the product is stable for the proposed shelf life of 18 months at +2 to +8 °C.

The CVMP recommended the submission of a variation application on removal of the thiomersal from the presentations after approval of marketing authorisation of CircoMax Myco. A commitment has been provided by the applicant.

The CVMP recommended the further identification of the source of the bioburden found during the manufacturing of the *M. hyopneumoniae* component by performing a thorough investigation/root cause analysis. The applicant has committed to provide the root-cause analysis.

The CVMP recommended the inclusion of the prolonged holding times after neutralisation and clarification, respectively, in the root-cause analyses to rule these out as the/a source of bioburden. In case of failure to rule out the holding times as a source of bioburden, the holding times should be reduced to a justified and validated level in order to reduce the risk of biomass multiplication and ensure a tolerable level of process consistency.

Part 3 – Safety

Introduction and general requirements

The active substances porcine circovirus PCV2a antigen (inactivated, recombinant) and *Mycoplasma hyopneumoniae* antigen (inactivated) of CircoMax Myco have been already authorised in the EU. However, PCV2b is a new antigen not included in a veterinary medicinal product in the EU before. A full safety file in accordance with Article 12(3)(j) has been provided.

Safety documentation

The applicant has provided target animal safety data based on laboratory studies with regard to single dose and split-dose administration and also based on field studies. A user risk assessment is also provided.

No data are provided on reproductive safety nor the effect on lactation and pregnancy as the vaccine is only indicated for the vaccination of piglets.

List of laboratory safety studies:

Study reference	*Study title*	*Batch used*
Repeated dose, 3-day-old piglets	B924N-NL-15-457	L1114LW05
Supportive data Single dose and repeated dose, 3-week-old pigs	B924R-US-17-732	L0517LW20
Supportive data Single dose and 10x overdose, 3-week-old pigs	B924R-JP-17-692	193755A
Supportive data 2x overdose 3-week-old pigs	B924R-US-15-524	L1014LW07 L0215LW06 L0215LW07
Supportive data 2x overdose 3-week-old pigs	B924R-US-17-733	L0517LW20 L0517LW21 L0517LW22

Laboratory tests

The applicant performed one good laboratory practice (GLP)-compliant study to assess single dose safety and split-dose administration safety with CircoMax Myco (study B924N-NL-15-457). Additionally, the applicant submitted the results of two safety studies performed for regulatory authorities outside the EU, study B924R-US-17-732 and study B924R-JP-17-692. These studies provide supportive safety data.

The vaccine batches used in the laboratory safety trials were manufactured according to the manufacturing process described in part 2.B of the dossier with some minor deviation not considered to adversely impact the safety of the product. The batch used in the GLP-compliant EU study was of maximum potency (4.9 [PCV2a], 5.9 [PCV2b] and 4.7 [*M. hyopneumoniae*] RP/dose).

The study represents the intended label vaccination schedule including administration of the product once according to the single dose scheme, and twice according to the split-dose scheme (D0 and D14).

Vaccinations neither caused systemic abnormal reactions nor did they adversely affect growth. Rectal temperature was not statistically different in vaccinated animals compared to negative controls, although transient temperature increases were observed in vaccinated piglets 4 hours and 1 day after vaccination. Injection site swellings of less than 2 cm in diameter occurred in the majority of piglets and lasted 9–10 days. Swellings were caused by an influx of inflammatory cells and considered a normal response to vaccination with an adjuvanted vaccine.

In summary, local and systemic adverse reactions such as redness and swelling at the injection site (below 2 cm in diameter for up to 10 days) and transient increase in body temperature during the first 24 hours after vaccination (not exceeding 2.1 °C) were seen and indicated as common and very common, respectively. Adverse reactions from this pivotal study were thus adequately reflected in the product literature.

Safety of one administration of an overdose

The demonstration of overdose safety is not required for inactivated vaccines anymore since Directive 2009/9/EC amending Directive 2001/82/EC came into force. Nevertheless, the applicant submitted the results of three safety studies which were performed for regulatory authorities outside the EU and did therefore not entirely comply with EU legislation and/or regulatory guidance. In two US studies, the safety of a single dose and of repeated administrations of single doses was assessed (B924R-US-15-524 and B924R-US-17-733). In the Japanese study, the safety of a single dose and a ten-times overdose was assessed in 3-week-old piglets (B924R-JP-17- 692).

Besides the reactions already mentioned in section 4.6, depression and reduced appetite were observed. The swellings were moderate (up to 7.5 cm diameter up to 13 days). Injection site reactions were found to be caused by local inflammatory response. Transient depression, reduced appetite, lameness and dyspnoea were also sporadically observed, but were not clearly related to vaccination. Supportive evidence from the overdose studies were included in section 4.10 of the SPC.

Safety of the repeated administration of one dose

The safety of two doses of vaccine given with 2 weeks interval was studied in study B924N-NL-15-457 described above. Local reactions and transient increase in body temperature were observed.

Examination of reproductive performance, lactation and pregnancy

No reproductive studies were provided as the product is only indicated for fattening animals. The safety of the veterinary medicinal product has not been established during pregnancy or lactation. The use of the product is not recommended during pregnancy and lactation.

Examination of immunological functions

A justification has been provided in this section of the dossier, but no specific trials have been carried out. It is not common for inactivated vaccines to have adverse immunological effects that affect the overall safety of the target species. As the laboratory data show that vaccine batches with maximum potency were well tolerated in seronegative pigs of minimum age, it is considered that no further examination of immunological functions is required.

User safety

The applicant provided a user safety risk assessment compliant with the CVMP 'Guideline for user safety for immunological veterinary products' (EMEA/CVMP/IWP/54533/2006). Minor potential risks were identified, which are essentially associated with handling and accidental self-injection. No special precautions need to be taken by the person administering the product to animals, as mentioned in section 4.5. of the SPC.

CircoMax Myco is an inactivated adjuvanted vaccine intended to be administered by professional users. The vaccine contains three components, two of which are chimeric PCV strains that have been specifically created using genetic modifications. The two PCV virus components and *Mycoplasma hyopneumoniae* do not infect humans and are inactivated in the product.

The adjuvant is composed of squalane (a naturally occurring material which is considered as not falling within the scope of Regulation No 470/2009). Although polysorbate 80 and poloxamer 401 can rarely induce adverse reactions in humans (hypersensitivity and anaphylactic-type reactions, respectively), none of the substances is included in the Annex to the European Commission guideline on 'Excipients in the labelling and package leaflet of medicinal products for human use' (EMA/CHMP/302620/2017 Rev. 1), and the risk is deemed to be minimal. Polysorbate 80 and poloxamer 401 (both included in Table 1 (allowed substances) of the annex to Commission Regulation (EU) No 37/2010). Squalane incorporated in non-mineral SP oil is also used in other porcine vaccines from the applicant, such as Suvaxyn Circo+MH RTU, which was centrally authorised in 2015 (EMEA/V/C/003924).

The preservative thiomersal is an allowed substance for which table 1 of the annex to Commission Regulation (EU) No 37/2010 indicates that no MRL is required. It is used at a concentration of 0.01% (w/v), which is below the maximum permitted concentration (0.02%) as stated in Commission Regulation (EU) No 37/2010.

Regarding gentamycin, the maximal theoretical concentration in the finished product would be well below biological activity levels. Gentamycin is used during the manufacturing process of both PCV2 components to reduce contamination risks and is found only in trace amounts in the finished product, thus not likely to pose a risk for the vaccine user.

Based on the above risk assessment, the CVMP concluded that the product does not pose an unacceptable risk to the user when used in accordance with the SPC.

Study of residues

No investigation into this aspect is required for this vaccine. The active ingredients being substances of biological origin intended to produce active immunity, do not fall within the scope of Regulation (EC) No 470/2009 with regard to residues of veterinary medicinal products in foodstuffs of animal origin. The vaccine does not contain adjuvants or any other excipients that require a numerical MRL or fall within the scope of Regulation (EU) No 470/2009 regarding residues of veterinary medicinal products in foodstuffs of

animal origin. The consumption of products derived from pigs vaccinated with the present IVMP does therefore not present a risk for consumer (human) health.

Gentamicin used in the cell culture-based production of the of the PCV-2 components is present at low residual levels in the finished medicinal product. Even in a worst case scenario, the maximum theoretical levels of gentamicin at injection site would still be approximately 10 times below the MRL established for porcine muscle.

Consequently, the withdrawal period in section 4.11 of the SPC is set at zero days.

Interactions

The applicant did not provide any safety studies investigating the concurrent use of the IVMP with any other veterinary medical product. Thus, section 4.8 of the SPC includes a standard warning.

Field studies (safety)

List of field safety studies:

Study reference	*Study title*	*Batch used*
Intermediate potency	B921C-NL-16-664, B921C-NL-16-643	193755D
Minimum potency	B826C-ES-16-661, B826C-ES-16-662, B826C-BE-16-663, B826C-ES-16-640, B826C-ES-16-641, B826C-BE-16-642	229878A
Supporting evidence	B921R-US-16-609	193748B/193676B

The applicant performed two field studies in The Netherlands, one for each dosing regimen (single dose B921C-NL-16-643 and split-dose B921C-NL-16-664), to address the safety of the IVMP under field conditions. These studies were performed with vaccine batches of intermediate potency (RPs of 2.5 [PCV2a], 3.9 [PCV2b], 2.9 [*M. hyopneumoniae*]), which is acceptable for field trials with veterinary vaccines.

The findings in the laboratory studies were confirmed by the field studies, as transient increases in mean rectal temperature were observed. The increase was within the range stated in the SPC: 'A transient increase in body temperature, not exceeding 2.1 °C, is very common during the first 24 hours after vaccination'. After single dose vaccination, transient local injection site reactions in 2 of the subsets of 15 vaccinated pigs (13.3%), consisting of a mild swelling of 0.5 to 2 cm in diameter with a maximum duration of 3 days. For the split-dose vaccination none of the piglets presented swellings at the injection site. No notable difference was observed concerning average daily weight gain and average body weight 30 days post-vaccination between control animals and vaccinated piglets. No abnormal clinical signs were observed in neither group after vaccination.

Additionally, the applicant submitted the results of six EU field efficacy studies performed with IVMP batches containing the active components at or near minimum potency and a US field safety study (supporting evidence, intermediate potency), involving 3-day-old pigs (split-dose vaccination schedule — 2 doses of 1 ml — 4 studies) and 3-week-old pigs (single dose vaccination schedule — 1 dose of 2 ml — 4 studies).

In summary, it is agreed that the results did support the safety data already generated in the laboratory studies and indicated that a single label dose of the vaccine was safe in 3-weeks-old piglets under field

conditions. Likewise, it was agreed that a split-dose of the vaccine was safe in 3-day- and 3-week-old piglets under field conditions.

Environmental risk assessment

Although immunological products do not require a phase I/II assessment as outlined by VICH GLs 6 and 38, the risks of vaccination with the IVMP to the environment were comprehensively described by the applicant in a phase I risk assessment. The overall risk of the vaccine to the environment, humans and other animals is effectively zero.

Environmental risk assessment for products containing or consisting of genetically modified organisms

Not applicable.

Overall conclusions on the safety documentation

The applicant provided satisfactory risk assessments for the user and the environment and included appropriate warnings and guidance in the SPC where necessary.

In summary, the applicant has provided data showing that the IVMP has a safety profile with mainly local reactions and transient temperature increases as possible adverse reactions. The vaccine is safe in minimum age piglets when administered by the recommended route and regimens. The maximum potencies and safety statements of the SPC are considered supported and appropriate.

Based on the data provided, it was concluded that target animal safety is acceptable when the vaccine is administered according to the recommended schedule and via the recommended route.

Reproduction safety was not investigated as the product is only intended for use in piglets.

A user safety assessment in line with the relevant guidance document has been presented.

Based on the assessment presented, the product does not pose an unacceptable risk to the user when used in accordance with the SPC.

The consumption of products derived from pigs vaccinated with the IVMP does not present a risk for consumer (human) health. Consequently, the withdrawal period in section 4.11 of the SPC is set at zero days.

CircoMax Myco is not expected to pose a risk for the environment when used according to the SPC.

The CVMP concluded that CircoMax Myco is not expected to pose a risk to the user or to the environment when used in accordance with the SPC.

Part 4 – Efficacy

Introduction and general requirements

The vaccine is intended for the active immunisation of piglets against porcine circovirus type 2 to reduce viral load in blood and lymphoid tissues, fecal shedding, the lesions in lymphoid tissues associated with PCV2 infection; and against *Mycoplasma hyopneumoniae* to reduce the lung lesions associated with *Mycoplasma hyopneumoniae* infection. Onset of immunity is intended to be established 3 weeks further to

the completion of either a single dose or split dose vaccination schedule and lasting for 23 weeks. Additionally, reduction on body weight gain losses under field conditions has been claimed by the applicant.

A specific Ph. Eur. monograph that outlines the requirements for inactivated PCV2 vaccines does not exist; therefore, efficacy parameters were selected by the applicant. Appropriate challenge models against PCV2a, PCV2b and PCV2d were established. For *Mycoplasma hyopneumoniae*, the requirements outlined in the specific Ph. Eur. monograph were followed (European Pharmacopoeia 8.0: Porcine enzootic pneumonia vaccine inactivated (04/2013:2448).

Efficacy was demonstrated in compliance with the European Directive 2001/82/EC (as amended by 2004/28/EC and Directive 2009/9/EC), and the European Pharmacopoeia (Ph. Eur.) chapter 5.2.7. "Evaluation of efficacy of veterinary vaccines and immunosera".

Challenge model:

PCV2 systemic disease is difficult to induce experimentally, and success depends on the inclusion of colostrum-deprived (CD) piglets, a challenge below three weeks of age and co-infection with other porcine pathogens. Not all these criteria can be controlled in a vaccine efficacy trial, therefore other efficacy parameters such as viral load in blood and/or tissues, viral shedding and characteristic histological tissue lesions in conjunction with the presence of PCV2 in those tissues were included. The applicant developed four challenge models, against PCV2a, PCV2b, PCV2d and *Mycoplasma hyopneumoniae*, respectively. The four models are summarised below.

Challenge model against Porcine Circovirus type 2a

- The challenge strain was PCV2, Isolate 40895 Passage 7. The PCV2a was passaged serially in porcine kidney cells (PK15) up to passage 6.
- Challenge volume of 3 ml: 1 ml administered intramuscular and 2 ml administered intranasally
- Evaluated parameters were viremia, faecal shedding, lymphoid tissue lesions and presence of antigen in lymphoid tissues
- Desired primary outcome was reduction of viremia; secondary outcomes were reduction of faecal shedding, reduction of lymphoid tissue lesions, and reduction of detection of PCV2 antigen within lymphoid tissue lesions.

Challenge model against Porcine Circovirus type 2b

- The challenge strain was PCV2, Isolate FD07 Pass 17. The PCV2b P16 was used to infect PK15cells. The day 9 and 12 harvests were combined to produce the challenge virus.
- Challenge volume of 3-4 ml: 1-2 ml administered intramuscular and 2 ml administered intranasally
- Evaluated parameters were viremia, faecal shedding, lymphoid tissue lesions and presence of antigen in lymphoid tissues
- Desired primary outcome was reduction of viremia; secondary outcomes were reduction of faecal shedding, reduction of lymphoid tissue lesions, and reduction of detection of PCV2 antigen within lymphoid tissue lesions

Challenge model against Porcine Circovirus type 2d

- The challenge strain was PCV2, Isolate PCV2d DIV P7 Lot# DS047-4-24Apr13. Challenge volume of 4 ml: 2 ml administered intramuscular and 2 ml administered intranasally

- Evaluated parameters were viremia, faecal shedding, lymphoid tissue lesions and presence of antigen in lymphoid tissues
- Desired primary outcome was reduction of viremia; secondary outcomes were reduction of faecal shedding, reduction of lymphoid tissue lesions, and reduction of detection of PCV2 antigen within lymphoid tissue lesions

Challenge Model against *Mycoplasma hyopneumoniae*

- The challenge strain was *Mycoplasma hyopneumoniae* strain 232 obtained from Ames, Iowa (2004). The lung homogenates were obtained after intra-tracheal inoculation of SPF pigs and collection of affected lung lobes to obtain a tissue homogenate. Challenge volume was 10 ml on one or two consecutive days, intra-tracheal or trans-tracheal route
- Evaluated parameters were development of macroscopic lung lesions and the primary outcome was reduction of macroscopic lung lesions.

The above challenge protocol to assess the efficacy of the product versus *Mycoplasma hyopneumoniae* was already accepted and used in another authorised product from the same company (Suvaxyn Circo+MH RTU). All challenge models were considered adequately validated and therefore appropriate for using in the efficacy trials in order to mimic the natural conditions for infection.

Efficacy parameters and tests:

The applicant chose viral load in blood (viremia), faecal shedding, lymphoid depletion and PCV2 colonisation of lymphoid tissue as efficacy parameters for the PCV2 component. Histiocytic replacement was also assessed, because it was a sequela to lymphoid depletion. Thus, in some studies the two parameters were analysed together to evaluate lymphoid lesions. The collection of blood and faeces allowed frequent sampling at multiple time points post challenge in order to study development of viremia and shedding over time.

Lung lesions were the primary end point for efficacy against *Mycoplasma hyopneumoniae* challenge.

Efficacy documentation

Studies included in part 4

The below long list of all laboratory and field (safety and) efficacy studies were conducted in support of this MA application and included studies both for the EU-dossier as well as for the US-dossier.

Overview of efficacy studies

Type of study	Dose	Challenge strain	Study number
Dose determination			
	Single dose	PCV2a	B823R-US-15-550
		PCV2b	B823R-US-15-551
		PCV2b	B823R-US-16-646
		M. hyopneumoniae	B823R-US-16-644
	Split dose	PCV2b	B823W-US-15-532
Onset of immunity			
	Single dose	PCV2a	B822R-US-14-325
		PCV2a	B822R-US-16-583
		PCV2b	B822R-US-16-582

		PCV2b	B822R-US-15-562
		PCV2d	B820R-US-17-747
		PCV2d	16PRGBIO-01-01
		M. hyopneumoniae	B822R-US-15-544
	Split dose	PCV2a	B822R-US-15-556
		PCV2a	B825R-US-16-667
		PCV2b	B822R-US-15-557
		PCV2d	B820R-US-17-747
		PCV2d	16PRGBIO-01-01
		M. hyopneumoniae	B822R-US-15-558
		M. hyopneumoniae	B822R-US-16-622
Effect of maternally derived immunity			
	Single dose	PCV2a	B828R-US-16-669
		PCV2b	B828R-ES-15-476
		PCV2b	B828R-ES-16-613
		M. hyopneumoniae	B828R-ES-15-474
	Split dose	PCV2a	B828R-ES-17-707
		PCV2a	B828R-US-18-797
		PCV2b	B823W-US-15-532
		M. hyopneumoniae	B822R-US-14-332
Duration of immunity			
	Single dose	PCV2a	B824R-US-15-452
		PCV2b	B824R-US-15-451
		PCV2d	B824R-US-19-890
		M. hyopneumoniae	B824R-US-17-751
	Split dose	PCV2a	B824R-GB-16-635
		PCV2b	B824R-GB-16-636
		M. hyopneumoniae	B824R-US-17-682
Field studies			
	Single dose		B826C-ES-16-640
			B826C-ES-16-641
			B826C-BE-16-642
	Split dose		B826C-ES-16-661
			B826C-ES-16-662
			B826C-BE-16-663

Dose determination

The predecessor product of CircoMax Myco is the porcine vaccine Suvaxyn Circo+MH RTU which was authorised by the centralised procedure in November 2015. CircoMax Myco contains the same MH and chimeric PCV2a master seeds as Suvaxyn Circo+MH RTU. The antigen inputs and potencies of the PCV2a component differ between the two vaccines. Additionally, CircoMax Myco contains a new chimeric PCV2b strain for which a new master seed was established. The chimeric PCV2b was essentially built the same way as the chimeric PCV2a, using the same PCV1 strain as the backbone for inclusion of a PCV2b ORF2 gene sequence.

The applicant submitted a total of five dose determination studies (one for PCV2a; two for PCV2b; one for MH) following single dose administration. Additionally, one dose determination study for the PCV2b

following the split dose administration (2x1 ml) was submitted. The studies showed that a potency of 1 RP for the PCV2 components was efficacious in MDA-negative piglets, even though higher potencies performed notably but not significantly better in these studies. However, study B823W-US.15-532 which included MDA-positive and -negative piglets, demonstrated that a potency of 1 RP was not sufficient to overcome interference by MDAs. Therefore, the minimum potency above 1 RP was recommended for the PCV2 components of CircoMax Myco. In the proposed SPC, a minimum RP is defined at 1.5 RP.

In study B823R-US-16-644 the applicant showed that a single dose of CircoMax Myco with MH potencies of RP ≥0.5 reduced lung lesions caused by a virulent MH challenge. The study was compliant with Ph. Eur. monograph 2448 requirements.

Onset of immunity

The applicant performed four challenge studies, two for each vaccination scheme, to determine the onset of immunity of the PCV2a component of CircoMax Myco at or below minimum potency.

Likewise, the applicant performed three challenge studies, to determine the onset of immunity of the PCV2b, including two studies with the single-dose scheme and one study with the split-dose scheme, at or below minimum potency. Additionally, the applicant submitted one immunogenicity study with a virulent PCV2d challenge.

Furthermore, the applicant performed three Ph. Eur. monograph 2448 compliant challenge studies to determine the onset of immunity of the *Mycoplasma hyopneumoniae* component of CircoMax Myco (one study with the single dose scheme and two studies to support the split dose scheme).

Results:

Colonisation of lymphoid tissue leading to lymphoid depletion and histiocytic replacement associated with PCV2a infection was more variable following challenge, such that statistical reductions in vaccinated pigs could not be shown in all studies. This would have required more animals to investigate statistical differences. Since numerically less vaccinated pigs than control pigs showed lesions in lymphoid tissue after challenge in all studies and significant differences were achieved in three out of four studies for lymphoid depletion and colonisation, then overall a reduction in viral loads and lesions in lymphoid tissues caused by PCV2a could be considered demonstrated.

Challenges with a heterologous PCV2b strain consistently induced viremia, fecal shedding and tissue lesions. Vaccination significantly reduced the amount of virus in blood and faeces as well as the frequency of viremic and shedding pigs. Vaccination also reduced the frequency of pigs with lymphoid tissue PCV2 colonisation and lesions. In conclusion, an overall reduction in viral loads and lesions in lymphoid tissues caused by PCV2b was demonstrated.

In one single-dose onset of immunity study (study B822R-US-16-582) including groups challenged with PCV2b, with two different vaccine batches, as well as a placebo and a negative control groups, significant differences could not be demonstrated for each parameter between vaccinated and both control groups because numerous piglets were excluded from the analysis due to sero-positivity, resulting in an underpowered study.

An additional immunogenicity study B820R-US-17-747 with a virulent PCV2d challenge demonstrated cross-protection afforded by Circomax Myco against viremia and fecal shedding caused by a PCV2d infection and confirmed the stimulation of cell-mediated immune responses against PCV2a, PCV2b and PCV2d. The onset of immunity against PCV2d was six weeks following a single dose and three weeks following a split dose scheme of CircoMax Myco.

A new study was submitted with the response to the list of questions (LOQ) supporting onset of immunity for cross-protection against PCV2d: Comparative study of three commercial *Mycoplasma hyopneumoniae* and Porcine Circovirus vaccine protocols in a dual challenge with *M. hyopneumoniae* and PCV2d (16PRGBIO-01-01)

The study was performed from September 2016 to April 2017 in the US. Briefly, 800 healthy pigs were used in this study. 192 pigs were randomly selected for the efficacy trial. Animals had no known history of vaccination or infection with *M. hyopneumoniae* or PCV2. 20 sows were bled 30 days before farrowing to determine PCV2 antibody levels of the herd. Pigs were individually identified on day 3 of age and randomly allocated to 8 treatment groups. Group 2 and 3 were immunized with CircoMax Myco. Group 2 once with 2 ml dose and group 3 with 2 doses of 1 ml (split dose vaccination scheme)

Animals were challenged with *M. hyopneumoniae* (LI-AI6 39 lung homogenate, 10 ml pre diluted, intratracheally) on day 53 of the study at 8 weeks of age (2 weeks after the second vaccination). On day 60, one week after the *M. hyopneumoniae* challenge, animals were challenged with 2 ml PCV2d intranasally and with 1 ml intramuscularly (cell culture propagated, $10^{4.5}$ to $10^{5.5}$ $TCID_{50}$). A subset of pigs (n=24) was necropsied 3 weeks (day 81 of the study) after PCV2d challenge. The remaining pigs were kept for another 173 days until they have reached 285 pounds to investigate pig performance.

In summary, results showed that groups 2 and 3, which were immunized with CircoMax Myco via single or split vaccination scheme, showed lower PCV2 viral load in blood, reduced virus shedding via faeces, reduced virus load in lungs compared to control group immunized with saline (group 1). No differences were observed for lung lesions or viral loads in other tissues.

These studies demonstrated protection against the subtypes 2a and 2b as well as cross-protection against subtype 2d and are thus considered acceptable for protection against the PCV2 subtypes 2a, 2b and 2d. The three challenge studies with heterologous *Mycoplasma hyopneumoniae* strains were considered valid, and vaccination consistently resulted in a significant reduction in lung lesions compared to control piglets.

It was concluded that vaccination with CircoMax Myco by the recommended route with doses as recommended in the SPC was efficacious and met efficacy requirements 3 weeks post vaccination.

Duration of immunity

Two duration of immunity studies were performed for each PCV fraction of the vaccine, and two duration of immunity studies were performed for the *Mycoplasma hyopneumoniae* fraction, that covered both vaccination schemes. Virulent challenges for both vaccine fractions (PCV and *Mycoplasma hyopneumoniae*) were performed approximately 23 weeks after vaccination corresponding to the normal production lifespan of commercial pigs.

Vaccine batches used in these studies contained the PCV2 fractions at a potency of approximately 1 RP and below the minimum potency. All challenges induced sufficient viremia and shedding in non-vaccinated control pigs. PCV2 tissue colonisation and lesions were less frequently observed in non-vaccinated control pigs. The reasons for this are not entirely clear. There is no specific Ph. Eur. monograph for PCV2 vaccines, and thus duration of immunity claims should be supported by evidence of protection according to general Ph. Eur. monograph 50207. There was less PCV2 viremia and virus fecal shedding for 23 weeks following either vaccination schemes. Protection in terms of reduced tissue colonisation and lesion scores was shown in studies where the challenge was virulent enough.

An additional US study performed in the field but with experimental challenge was submitted with the response to the LOQ in support of duration of immunity: Field duration of immunity 24 weeks after intramuscular administration of the porcine circovirus vaccine, Type 1- Type 2 chimera, killed virus, product code 19K5.R1, against a porcine circovirus type 2d challenge (B824R-US-19-890).

This study was performed in April 2019 to October 2019. The objective of the study was to demonstrate field duration of immunity of at least 24 weeks.

Briefly, 160 animals were allocated to 4 groups (n=40). The groups of animals were immunized once on day 0 of the study with a 2 ml dose of PCV2a2b (groups 2 to 4, different batches, 237330A, 280437F and 282997D) intramuscularly into the right neck and one group served as a control (group 1, saline control). Challenge infection was performed with PCV2d, isolate Z12 (Stock Lot# 082218PCVd) at day 167 with a total of 6 ml. 4 ml were applied intranasal and 2 ml were applied intramuscularly into the left neck. Animals were necropsied on day 187/188 of the study.

In summary, the study results showed that the claims on reduction in viral loads in blood and virus faecal shedding were fulfilled. For lymphoid depletion the results were also found significant but not for histocytic replacement between vaccinated and control groups.

Vaccine batches used in the *Mycoplasma hyopneumoniae* studies contained a potency of approximately 1.5 RP, which is the minimum proposed potency. Post challenge lung lesions were observed in non-vaccinated control pigs and a significant reduction in lesions could be demonstrated in vaccinated pigs following the single dose vaccination scheme. The DoI study on the split dose vaccination scheme for *Mycoplasma hyopneumoniae* was significantly confounded by infection in the experimental pigs with growth of secondary bacteria from lung swabs collected at necropsy. Therefore, the proposed DoI was supported by data from an already authorised vaccine (Suvaxyn Circo + MH RTU) with the same *M. hyopneumoniae* antigen included at a relative potency of 1.5 - 3.8 RP. In that study a duration of immunity was demonstrated to be 23 weeks. Although the reference vaccines used to potency test the MHyo fraction for the two vaccines are not the same, the reference vaccine used to potency test the MHyo fraction of CircoMax Myco is more potent than the corresponding reference for Suvaxyn Circo+MH RTU. Study results from the split-dose vaccination schedule show comparable efficacy to the single dose schedule in the present dossier, and in the latter schedule a DoI on 23 weeks was demonstrated. Also, no interference of the PCV2 antigens was demonstrated on the efficacy of the *M. hyopneumoniae* fraction, therefore convincing data from the Suvaxyn Circo + MH RTU dossier also were considered relevant for the present CircoMax Myco application. For both vaccination schedules, a duration of immunity of 23 weeks thereby was accepted.

Maternally derived antibodies (MDA)

Three MDA studies were performed in MDA positive piglets to assess the efficacy of the PCV2a component of CircoMax Myco, with an additional three MDA studies for the PCV2b component, and two MDA studies for the *Mycoplasma hyopneumoniae* component, according to recommendations from the EMA reflection paper describing the demonstration of MDAs on vaccine efficacy.

For the PCV2a studies, one study with the single dose vaccination scheme (B828R-US-16-669) confirmed reduced viremia and fecal shedding when administered to MDA positive piglets. Infection and destruction of lymphoid tissue was also reduced, although a significant difference was not shown because the challenge was not virulent enough to induce sufficient tissue lesions. A second study with MDA positive piglets using the split dose vaccination scheme (B828R-ES-17-707) was invalid because the PCV2a challenge was not virulent enough and the study was underpowered. This study could only be regarded as supportive. In the repeated split dose study (B828R-US-18-797), protection against viremia, shedding, lymphoid infection and tissue lesions caused by PCV2a could be demonstrated in MDA positive pigs following vaccination.

For the PCV2b studies, results of the first study with the single dose vaccination scheme (B828R-ES-15-476) did not show any protective effect of vaccination in the presence of MDAs. MDA titres before vaccination were high in this study because dams were vaccinated during pregnancy. A repeated study

(B828R-ES-16-613) with piglets that had moderate MDA levels between 0.5 and 1.3 S/P demonstrated a significant reduction in viremia and lymphoid colonisation and a numerical reduction in fecal shedding, lymphoid depletion and histiocytic replacement caused by PCV2b. Thus, vaccination could overcome interference of MDAs as long as MDA levels were not high. Consequently, an SPC warning is included in section 4.9 to postpone vaccination when very high MDA levels are expected. A third study was performed using the split dose scheme (B823W-US-15-532) and showed, that PCV2b potencies of >1 RP were required to overcome the adverse effects of MDAs, as already discussed in the dose determination section. An additional SPC recommendation is included in section 4.9 of the SPC to use the split dose vaccination scheme in piglets with moderately high levels of MDAs. The two vaccination schemes were never compared in MDA positive piglets in the same study design. Comparing the efficacy results of all MDA studies demonstrated that the split dose vaccination scheme provided notably better protection against PCV2 infection than the single dose scheme in the presence of MDAs.

For the *Mycoplasma hyopneumoniae* component, one study using the single dose vaccination scheme (B828R-ES-15-474) confirmed that vaccination with CircoMax Myco reduced lung lesions caused by *Mycoplasma hyopneumoniae* when administered in the presence of MDAs. The results of a second study (B828R-US-14-332) with MDA positive piglets and Suvaxyn PCV2bMH, the smaller combination fall-out of CircoMax Myco containing only PCV2b as PCV component, administered by the split dose scheme also demonstrated protection from lung lesions. The results were accepted as supportive data in accordance with the EMA guideline on combined vaccines (EMA/CVMP/IWP/594618/2010) which states that it is possible to support the efficacy of the larger combination with the results from a challenge study with a vaccine containing fewer active substances. No adverse effects of MDAs on the efficacy of the *Mycoplasma hyopneumoniae* component of CircoMax Myco were observed in these studies.

It was concluded that vaccination by the recommended route with doses of the minimum content recommended in the SPC were efficacious and met the Ph. Eur. efficacy standard including MDA positive animals.

Interactions

No information is available on the safety and efficacy of this vaccine when used with any other veterinary medicinal product. A decision to use this vaccine before or after any other veterinary medicinal product therefore needs to be made on a case by case basis.

Field trials

In order to assess the efficacy of CircoMax Myco in field conditions, six studies were conducted in two different EU countries and involving 3-day-old pigs (split dose: 3 studies) and 3-week-old pigs (single dose: 3 studies). The studies conducted in Spain were designed to confirm the claims for PCV2; protocol amendments reflect the applicant's decision to change the intended claim for this vaccine to PCV2-related claims only and not PCV2 genotype specific claims. The studies conducted in Belgium were designed to confirm the claims for *M. hyopneumoniae*.

Study reference	Study title	Batch used
B826C-ES-16-640	Evaluation of the safety and efficacy of a Porcine circovirus Type 1-Type 2 chimera, killed virus - *Mycoplasma hyopneumoniae* vaccine when administered to piglets at 3 weeks of age under field conditions in Spain.	229878A

Study reference	Study title	Batch used
B826C-ES-16-641	Evaluation of the safety and efficacy of a Porcine circovirus Type 1-Type 2 chimera, killed virus - *Mycoplasma hyopneumoniae* vaccine when administered to piglets at 3 weeks of age under field conditions in Spain.	
B826C-BE-16-642	Evaluation of the safety and efficacy of a Porcine circovirus Type 1-Type 2 chimera, killed virus - *Mycoplasma hyopneumoniae* vaccine when administered to piglets at 3 weeks of age under field conditions in Belgium.	
B826C-ES-16-661	Evaluation of the safety and efficacy of the flexible dose of a Porcine circovirus Type 1-Type 2 chimera, killed virus - *Mycoplasma hyopneumoniae* vaccine when administered to piglets at 3 days and 3 weeks of age under field conditions in Spain.	
B826C-ES-16-662	Evaluation of the safety and efficacy of the flexible dose of a Porcine circovirus Type 1-Type 2 chimera, killed virus - *Mycoplasma hyopneumoniae* vaccine when administered to piglets at 3 days and 3 weeks of age under field conditions in Spain.	
B826C-BE-16-663	Evaluation of the safety and efficacy of the flexible dose of a Porcine circovirus Type 1-Type 2 chimera, killed virus - *Mycoplasma hyopneumoniae* vaccine when administered to piglets at 3 days and 3 weeks of age under field conditions in Belgium.	

The same vaccine batch no.: 229878A containing antigens at (or close to) minimum levels (PCV2a at 1.5 RP, PCV2b at 1.7 RP and MH at 1.5 RP) was used in all six studies.

The study design of the field studies was essentially the same and is summarised below. All six studies were randomised, negatively controlled, good clinical practice (GCP) compliant studies aimed to assess the safety and efficacy of the 'single dose' and 'split dose' vaccination schemes under field conditions. Studies were performed at two different locations in Spain and in a farm in Belgium.

In order for a farm to be suitable for this study it had to meet the following requirements:

- Farrow-to-finish, or finishers within control of investigator at different but nearby location
- Documented infection with PCV2
- Current problems with PCVD or history of PCVD in the last two years
- Non-vaccinating against PCV2 in sows for at least 6 months
- Non-vaccinating against *M. hyopneumoniae* in sows for at least 6 months
- Presence of *M. hyopneumoniae* lung lesions at slaughterhouse in >10% of the animals
- Documented infection with *M. hyopneumoniae.*

Post inclusion removal criteria: A serious disease that might interfere with the vaccination. Receipt of any medication that might interact with vaccination.

Observation period: From study day -1 until slaughter

The primary efficacy variable to assess efficacy against PCV2 was

- viremia and fecal shedding as determined via qPCR.

Secondary efficacy variables were

- *M. hyopneumoniae* lung lesion score at slaughter
- presence of PCV2 DNA (qPCR) in fecal swabs
- serology (ELISA) for PCV2 and *M. hyopneumoniae*
- average daily weight gain
- mortality
- histopathology for PCVD-like lesions and
- PCV2-specific immunohistochemistry (IHC) in lymphoid tissues.

The primary efficacy parameter to assess efficacy against *Mycoplasma hyopneumoniae* was lung lesions.

Results

An overview of the main results is given in the table below:

	Single dose			**Split dose**		
Study site	ES B640	ES B641	BE B642	ES B661	ES B662	BE B663
Mortality % (Con vs Vac)	9.6_9.3	23.2_21.6	6.5_6.1	12.5_12.1	36.2_32.1	9.6_7.6
Mortality difference	ns	ns	ns	ns	ns	ns
Body Weight gain	ns	+	ns	+	ns	ns
PCV parameters						
PCV strains detected	2b	2a 2d	2a	2b 2d	2a	2a
Viremia	+	+	+	+	+	+
Faecal shedding %	ns	ns	ns	ns	ns	ns
Faecal shedding amount	+ 5 mpv	+ ≥3 mpv	ns	+ ≥4 mpv	+ ≥3 mpv	ns
PCV2 colonization	+	+	ns	+	ns	ns
Lymphoid depletion	(ns)	ns	ns	ns	+	ns
Histiocytic replacement	+	+	ns	ns	ns	ns
Systemic PCV2 disease	ns	ns	-	ns	ns	-
Subclinical PCV2 infection	+	+	-	+	ns	-
Mycoplasma parameters						
MH serology	vac+	-	ns	vac+		vac+
MH lung lesions	+	ns	ns	ns	ns	ns
Legend	+	Significant difference in favour of the vaccinated group				
	ns	No significant difference				
	(ns)	Borderline significant difference				
	-	No reactions observed in neither group				
	mpv	months post vaccination				

In general, no significant differences between vaccinated and non-vaccinated groups with regard to mortality were observed in any of the studies.

The efficacy claims regarding PCV2- infection were generally supported by the field efficacy data. Thus, all study sites confirmed an effect of vaccination against PCV viraemia. At the two study sites in Spain the results showed effect of vaccination on faecal shedding (amount), PCV2 colonisation, histiocytic replacement and subclinical disease. The natural challenge at the study site in Belgium was apparently lower and no effect was seen on the secondary efficacy parameters.

Less clear results were seen for *Mycoplasma hyopneumoniae* under field conditions. The only efficacy parameter was the extent of lung lesions, and here only one study showed a statistically significant degree of protection.

In conclusion the field studies consistently confirmed the protective effect of the PCV2 viraemia and for the secondary PCV indicators for subclinical disease. The positive effect was seen both under the single dose and the split-dose vaccination schemes. The field studies provided some support for the protection of CircoMax Myco against lung lesions caused by *Mycoplasma hyopneumoniae* although a statistically significant effect was only seen in a single study.

The field studies provided evidence on reduction of the disease-induced losses of body weight, although not consistently. Two of the six study herds (one under the single dose regimen, and one under the split dose regimen) showed a statistically significant positive effect on body weight. Some reservation remains on one of the study sites (B826C-ES-16-641) as the farm was not able to account for all pigs in the experiment and as the mortality rate was unusually high.

A revised wording was proposed for the SPC claim, and agreed by the applicant: "In addition, vaccination has been shown to reduce body weight gain losses under field conditions"

Overall conclusion on efficacy

The laboratory studies have generally supported the 3 weeks claim for onset of immunity against PCV2a, PCV2b and *Mycoplasma hyopneumoniae* after the completion of either the single-dose or the split-dose vaccination schemes.

With regard to duration of immunity, the 23 weeks claim was supported for PCV2a, PCV2b and *Mycoplasma hyopneumoniae* components for the single dose vaccination scheme. However, for the split-dose vaccination scheme, efficacy could only be demonstrated experimentally for the PCV2a and PCV2b components. Efficacy against *Mycoplasma hyopneumoniae* after split dose vaccination was accepted based on evidence from the authorised vaccine Suvaxyn Circo + MH RTU with the same mycoplasma antigen, the higher corresponding potency specifications of the present product, the general non-inferiority of the split-dose vaccine in the experimental studies and background immunological arguments.

The field studies generally supported the claims of protection against PCV2a and PCV2b, and partly supported the claim for protection against *Mycoplasma hyopneumoniae* as statistically significant differences in lung lesions could only be confirmed in one of the six studies. Cross-protection against PCV 2d was demonstrated in three studies conducted in the USA.

Comparing the efficacy results of all MDA studies demonstrates that the split dose vaccination scheme provides notably better protection against PCV2 infection than the single dose scheme in the presence of MDAs. No detrimental effect of MDAs on the efficacy of the *Mycoplasma hyopneumoniae* component of CircoMax Myco were observed in relevant studies.

The field studies provided evidence on reduction of the disease-induced losses of body weight, although not consistently. Two of the six study herds showed a statistically significant positive effect on body weight.

Part 5 – Benefit-risk assessment

Introduction

CircoMax Myco is a vaccine containing porcine circovirus antigen (inactivated, recombinant) and *Mycoplasma hyopneum*oniae antigen (inactivated). The circovirus component is based on chimeric virus strains (PCV type 1/2a and PCV type 1/2b, respectively).

The product was initially indicated for the 'Active immunisation of pigs against porcine circovirus type 2 to reduce viral load in blood and lymphoid tissues, virus faecal shedding and the lesions in lymphoid tissues associated with PCV2 infection. To reduce body weight gain losses in finishers at slaughter in cases of early PCV2 viremia (before 16 weeks of age). Active immunisation of pigs against *Mycoplasma hyopneumoniae* to reduce the lung lesions associated with *Mycoplasma hyopneumoniae* infection'.

A revised list of indications is suggested by the CVMP:

Active immunisation of pigs against porcine circovirus type 2 to reduce viral load in blood and lymphoid tissues, fecal shedding and the lesions in lymphoid tissues associated with PCV2 infection. Protection was demonstrated against porcine circovirus types 2a, 2b and 2d.

Active immunisation of pigs against *Mycoplasma hyopneumoniae* to reduce the lung lesions associated with *Mycoplasma hyopneumoniae* infection.

Onset of immunity (both vaccination schedules): 3 weeks after (the last) vaccination.

Duration of immunity (both vaccination schedules): 23 weeks after (the last) vaccination.

In addition, vaccination has been shown to reduce body weight gain losses under field conditions

The dossier was submitted in line with requirements of Article 12(3) of Directive 2001/82/EC.

Benefit assessment

Direct therapeutic benefit

In well-conducted laboratory studies, the vaccine was shown to induce active immunisation of pigs against PCV2 subtypes 2a and 2b and reduce viral load in blood and lymphoid tissues and faecal shedding caused by infection with PCV2; and to induce active immunisation against *Mycoplasma hyopneumoniae* and reduce lung lesions. Cross-protection against PCV 2d was demonstrated in three studies conducted in the USA.

The product was shown to have an OOI at 3 weeks after vaccination with DOI of 23 weeks after vaccination against PCV2.

The OOI for *Mycoplasma hyopneumoniae* was likewise 3 weeks, and duration of immunity could be set at 23 weeks.

The efficacy of the vaccine was adequately confirmed in the presence of MDA.

The applicant proposed a claim for reduction of body weight losses, but this was not consistently confirmed in the field studies. A revised wording was proposed for the SPC section 4.2 and accepted by the applicant.

Additional benefits

Not applicable.

Risk assessment

Quality:

Overall, information on the development, manufacture and control of the active substances and the finished product has been presented in a satisfactory manner.

The manufacture of *M. hyopneumoniae* antigen is based on a seed lot system, a standard aerobic fermentation in bioreactors, followed by inactivation, purification and sterilisation by filtration. The manufacturing processes of the two PCV antigens are identical and comprise standard virus production in pig kidney PK-15 cells followed by separation of micro carriers and cell material, and concentration, purification, and inactivation of virus. The manufacturing process is based on a seed lot system. Generally, the manufacturing processes are considered adequately controlled. Based on the data from six consecutive finished product batches, acceptable batch-to-batch consistency is considered demonstrated. Compliance with Ph. Eur. monographs 0062 *Vaccines for veterinary use* is generally considered demonstrated.

Data from stability studies for six batches of the finished product indicate that the product is stable for the proposed shelf life of 18 months at +2 to +8 °C.

Three recommendations have been proposed by the CVMP:

The CVMP recommended the submission of a variation application on removal of the thiomersal from the presentations after approval of marketing authorisation of CircoMax Myco. A commitment has been provided by the applicant.

The CVMP recommended the further identification of the source of the bioburden found during the manufacturing of the *M. hyopneumoniae* component by performing a thorough investigation/root cause analysis. The applicant has committed to provide the root-cause analysis.

The CVMP recommended the inclusion of the prolonged holding times after neutralisation and clarification, respectively, in the root-cause analyses to rule these out as the/a source of bioburden. In case of failure to rule out the holding times as a source of bioburden, the holding times should be reduced to a justified and validated level in order to reduce the risk of biomass multiplication and ensure a tolerable level of process consistency.

Safety:

The applicant provided satisfactory risk assessments for the user and the environment and included appropriate warnings and guidance in the SPC where necessary.

In summary, the applicant has provided data showing that the IVMP has a safety profile with mainly local reactions and transient temperature increases as possible adverse reactions. The vaccine is safe in minimum age piglets when administered by the recommended route and regimens. The maximum potencies and safety statements of the SPC are considered supported and appropriate:

Based on the results it was concluded that the safety of the target animals was acceptable when the vaccine is administered according to the recommended schedule and via the recommended route.

Reproduction safety was not investigated as the product is only intended for piglets.

A user safety assessment in line with the relevant guidance document was presented. Based on that assessment, the potential health risk of the product to all users is considered low and acceptable when used in accordance with the SPC.

CircoMax Myco is not expected to pose a risk for the environment when used according to the SPC.

Risk management or mitigation measures

Appropriate information has been included in the SPC to inform on the potential risks of this product relevant to the target animal, user, consumer, and the environment and to provide advice on how to prevent or reduce these risks.

Evaluation of the benefit-risk balance

The product has been shown to be efficacious to reduce viral load in blood and lymphoid tissues, virus faecal shedding and the lesions in lymphoid tissues associated with PCV2 infection, and to induce active immunisation against *Mycoplasma hyopneumoniae* and reduce lung lesions.

The applicant proposed a claim for reduction of body weight losses, but this was not consistently confirmed in the field studies. A wording has been proposed for the SPC point 4.2 to reflect the actual findings of the field studies and has been accepted by the applicant.

With regard to quality, the CVMP has made two recommendations relating to the microbial load observed during the production of the *M. hyopneumoniae* antigen and one recommendation to remove thiomersal from the formulation. The applicant has made commitments to solve the concern.

CircoMax Myco is well tolerated by the target animals and presents an acceptable risk for users and the environment when used as recommended. The withdrawal period is set at zero days.

Conclusion

Based on the original and complementary data presented on quality, safety and efficacy the Committee for Medicinal Products for Veterinary Use (CVMP) concluded that the application for CircoMax Myco is approvable since these data satisfy the requirements for an authorisation set out in the legislation (Regulation (EC) No 726/2004 in conjunction with Directive 2001/82/EC).

The CVMP considers by consensus that the benefit-risk balance is positive and, therefore, recommends the granting of the marketing authorisation for the above mentioned veterinary medicinal product.

In addition, it is recommended that the applicant provides the following information, following approval of the marketing authorisation application:

- The CVMP recommended the submission of a variation application on removal of the thiomersal from the presentations after approval of marketing authorisation of CircoMax Myco. A commitment has been provided by the applicant.
- The CVMP recommended the further identification of the source of the bioburden found during the manufacturing of the *M. hyopneumoniae* component by performing a thorough investigation/root cause analysis. The applicant has committed to provide the root-cause analysis.

- The CVMP recommended the inclusion of the prolonged holding times after neutralisation and clarification, respectively, in the root-cause analyses to rule these out as the/a source of bioburden. In case of failure to rule out the holding times as a source of bioburden, the holding times should be reduced to a justified and validated level in order to reduce the risk of biomass multiplication and ensure a tolerable level of process consistency.